**Honors Bio Lab: The effect of substrate concentration on enzyme activity**

1. **Question:** How does substrate (H2O2) concentration affect enzyme (catalase) activity?
2. **Background info**

The enzyme catalase is found in many organisms, from celery to humans to yeast, because it plays a very important role in protecting cells. Hydrogen peroxide (H2O2) is a normal byproduct of cellular metabolism but it is toxic to cells – if it was not broken down it would kill cells. The enzyme **catalase speeds up the breakdown of hydrogen peroxide (H2O2) into water (H2O) and oxygen (O2)**, both of which are not harmful cells.

2 H2O2  2 H2O+ O2

Catalase

(substrate) (products)

1. Identify the following:

IV:

DV:

Control:

Constants:

Hypothesis:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

1. **Experiment**

Materials

* Small plastic cups (labeled)
* 3% H2O2
* Water
* Yeast suspension
* Forceps
* Plastic transfer pipettes
* Microwell plate
* Small filter disk

Procedure

1. Practice pipetting (no air bubbles)

2. Determine maximum amount of water that will fit in well

3. Fill wells will different dilutions of H2O2

|  |  |  |  |
| --- | --- | --- | --- |
| “Control”  2 mL ofH2O |  |  |  |
| “A”  1.5 mL ofH2O  0.5 mL ofH2O2 | “B”  1 mL ofH2O  1 mL ofH2O2 | “C”  0.5 mL of H2O  1.5 mL ofH2O2 | “D”  2 mL ofH2O2 |
|  |  |  |  |

4. Create the following data table with title in your notebook:

**Table 1: Time for yeast-soaked disk to float**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Control | A | B | C | D |
| Trial 1 |  |  |  |  |  |
| Trial 2 |  |  |  |  |  |
| Trial 3 |  |  |  |  |  |
| Average |  |  |  |  |  |

5. Gently stir/shake yeast suspension.

6. Using forceps pick up a filter disk and submerge it in the yeast suspension.

7. Be ready with timer. As soon as place the yeast-soaked disk in well start timing.

8. Record the time when the disk is all the way floating on top of the liquid. Record in SECONDS! If 120 seconds (two minutes) pass and the paper still isn’t floating just record “no rxn.”

9. Calculate the average time for each of the five different conditions.

1. **Analysis**
2. Graph the averages.
3. Which concentration of H2O2 yielded the fastest reaction time? \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_
4. Which concentration of H2O2 yielded the slowest reaction time? \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_
5. **Conclusion**
6. Does the data support or not support the hypothesis? EXPLAIN
7. What trends/patterns did you observe regarding the reaction times? Or maybe you observed no trends?
8. Why do you think you got the results you did?
9. Discuss possible sources of error.